The nucleotide sequence of the lac operon and phage junction in lambda gt11

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DESCRIPTION:The sequence of an EcoRI-BstNI fragment from lambda gt11 was determined using Maxam and Gilbert method. This sequence connects the lac operon and lambda sequences (1,2) and matches with the respective regions. The phage left arm sequence terminates at 110bp. The lambda-lac5 junction is indicated (|).

COMMENTS: The lom region of lambda and lac operon are transcribed in opposite directions in gt11 (2,3). Lambda gt11 was designed to express inserts as beta galactosidase fusion products (3). But in at least two instances (4,5) antigenic proteins were expressed even when the inserts were in the opposite orientation and were not fused to beta galactosidase. In the direction opposite to that of lac translation, the sequence shown here has one translation reading frame which could continue past the Eco site into the insert. The other reading frames including lom terminate prematurely. This open reading frame (ORF) starts with a met codon at 138bp and has a putative ribosome binding site (RBS). This ORF may be responsible for the expression of inserts in the opposite orientation, else translation would have to initiate within the insert. The sequence TACAAT at 100bp is like fd111 promoter (6) with a CACA sequence as a -35 region. Either this or some other upstream promoter on the l-strand might be responsible for transcription.

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lom ---->  .         .         .         .         .         .         .         .
ACGTTATGTGAGCGTGATGGCCGGACCGGTTTTACAAATCAGTAAGCAGGTCAGTGCGTACGCCATGGCCCGAGTGGCTCACAGTCGGTG 1  90
GCGAATACACTCGCACTACCGGCCTGGCCAAAATGTTTAGTCATTCGTCCAGTCACGCATGCGGTACCGGCCTCACCGAGTGTCAGCCAC r
            phage | lac 3'end   RBS          ------> Open Reading Frame
GTCCGGCAGTACAATGGATTTCCTTACGCGAAATACGGGCAGACATGGCCTGCCCCGTTATTATTATTTTTGACACCAGACCAACTGGTA 1 180
CAGGCCGTCATGTTACCTAAAGGAATGCGCTTTATGCCCGTCTGTACCGGACGGGCCAATAATAATAAAAACTGTGGTCTGGTTGACCAT r
                      EcoR I
ATGGTAGCGACCGGCGCTCAGCTGGAATTC 1 210                Translation/beta gal.fusion products
TACCATCGCTGGCCGCGAGTCGACCTTAAG r /Insert/EcoRI/lac/ <----------
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